Amendments to the Specification

Please replace the paragraph beginning at page 6, line 12, with the following rewritten paragraph:

--Figures 7A, 7B, and 7C show the construction of derivative PKS gene clusters from the vector of Figure 3.--

Please replace the paragraph beginning at page 6, line 14, with the following rewritten paragraph:

--Figures 8A-8B show antibiotics obtained from the polyketides of Figure 6A-6F.--

Please replace the paragraph beginning at page 28, line 10, with the following rewritten paragraph:

--For each of the six modules of DEBS, a subclone was made containing endonuclease restriction sites engineered at selected boundaries of the acyltransferase (AT) and reduction (KR or DH/ER/KR) domains. The restriction sites were introduced into the subclones by PCR mutagenesis. A *Bam*HI site was used for the 5' boundary of AT domains, a *Pst*I site was introduced between the AT and reductive domains, and *Xba*I was used at the 3' end of the reductive domain (see Figure 5). This resulted in the following engineered sequences (lowercase indicates engineered restriction site) (SEQ ID NOS:1-18):

Module 1 (pKOS011-16)

5' AT boundary	GCGCAGCAG*ggatcc*GTCTTCGTC
AT/KR boundary	CGCGTCTGG*ctgcag*CCGAAGCCG
3' KR boundary	CCGGCCGAA*tctaga*GTGGGCGCG

Module 2 (pKOS001-11)

5' AT boundary	TCCGACGGT*ggatcc*GTGTTCGTC
AT/KR boundary	CGGTTCTGG*ctgcag*CCGGACCGC
3' KR boundary	ACGGAGAGC*tctaga*GACCGGCTG

Module 3 (pKOS024-2)

5' AT boundary	GACGGGCGC*ggatcc*GTCTTCCTG
AT/KR boundary	CGCTACTGG*ctgcag*CCCGCCGCA
3' KR boundary	ACCGGCGAG*tctaga*CAACGGCTC

Module 4 (pKOS024-3)

5' AT boundary	GCGCCGCGC*ggatcc*GTCCTGGTC
AT(DH/ER/KR) boundary	CGCTTCTGG*ctgcag*CCGCACCGG
3' DH/ER/KR boundary	GGGCCGAAC*tctaga*GACCGGCTC

Module 5 (pKOS006-182)

5' AT boundary	ACTCGCCGC*ggatcc*GCGATGGTG
AT/KR boundary	CGGTACTGG*ctgcag*ATCCCCACC
3' KR boundary	GAGGAGGGC*tctaga*CTCGCCCAG

Module 6 (pKOS015-52)

5' AT boundary	TCCGCCGGC*ggatcc*GTTTTCGTC
AT/KR boundary	CGGTACTGG*ctgcag*CCGGAGGTG
3' KR boundary	GTGGGGGCC*tctaga*GCGGTGCAG--

Please replace the paragraph beginning at page 29, line 18, with the following rewritten paragraph:

-- A cosmid library of genomic DNA from *Streptomyces hygroscopicus* ATCC 29253 was used to prepare DNA cassettes prepared from the rapamycin PKS gene cluster to be used as replacements into the enzymatic activity regions of the erythromycin gene cluster. Cassettes were prepared by PCR amplification from appropriate cosmids or subclones using the primer pairs listed in Table 1, and were designed to introduce suitable restriction sites at the ends of the cassettes. The rapAT2 cassette is flanked by *Bgl*II and *Pst*I sites, and the rapAT14 cassette is flanked by *Bam*HI and *Pst*I sites. The reductive cycle cassettes are flanked by *Pst*I and *Xba*I sites. Large DH/ER/KR cassettes were amplified in two pieces, then joined at an engineered *Xho*I site in order to minimize errors introduced during PCR amplification of long DNA sequences. The rapKR4 cassette was made by cloning a 1.3kb *NheI*/*Xba*I fragment from the rapDH/KR4 cassette above into the *Xba*I site in pUC19. There is a *Pst*I site that is in-frame and upstream of *Xba*I in pUC19 that generates the following junction at the 5'-end of the cassette:

5'-*ctgcag*GTCGACTCTAGCCTGGT...(SEQ. ID NO. 19)--

Please replace the paragraph beginning at page 30, line 1, with the following rewritten paragraph:

Table 1 Primer pairs used for PCR amplification of rapamycin PKS cassettes. All primers are listed from 5' to 3'. Engineered restriction sites are lower case.		
Module	**Primer**	**Sequence (SEQ ID NOS:20-31)**
rapAT2	forward:	TTT*agatct*GTGTTCGTCTTCCCGGGT
	Reverse:	TTT*ctgcag*CCAGTACCGCTGGTGCTGGAAGGCGTA
rapAT14	Forward:	TTT*ggatcc*GCCTTCCTGTTCGACGGGCAAGGC
	Reverse:	TTT*ctgcag*CCAGTAGGACTGGTGCTGGAACGG
rapKR2	Forward:	TTT*ctgcag*GAGGGCACGGACCGGGCGACTGCGGGT
	Reverse:	TTT*tctaga*ACCGGCGGCAGCGGCCCGCCGAGCAAT
rapDH/KR4	Forward:	TT*ctgcag*AGCGTGGACCGGGCGGCT
	Reverse:	TTT*tctaga*GTCACCGGTAGAGGCGGCCCT
rapDH/ER/KR1	Forward:	TTT*ctgcag*GGCGTGGACCGGGCGGCTGCC
(left half)	Reverse:	TTT*ctcgag*CACCACGCCCGCAGCCTCACC
rapDH/ER/KR1	Forward:	TTT*ctcgag*GTCGGTCCGGAGGTCCAGGAT
(right half)	Reverse:	TTT*tctaga*ATCACCGGTAGAAGCAGCCCG

Please replace the paragraph beginning at page 30, line 4, with the following rewritten paragraph:

--The following are typical procedures. The products are indicated by their numbers in Figure 6, where "a" represents the embodiment where R is methyl; "b" represents the embodiment where R is hydrogen.

a) Replacement of DEBS DH/ER/KR4. A portion of the erythromycin gene of module 4 (eryDH/ER/KR4) was replaced either with the corresponding rapamycin activities of the first rapamycin module (rapDH/ER/KR1) or of module 4 of rapamycin (rapDH/KR4). The replacement utilized the technique of Kao *et al. Science* (1994) 265:509-512. A donor plasmid was prepared by first amplifying 1 kbp regions flanking the DH/ER/KR4 of DEBS to contain a *Pst*I site at the 3' end of the left flank and an *Xba*I site at the 5' end of the right flank. The fragments were ligated into a temperature-sensitive donor plasmid, in a manner analogous to that set forth for KR6 in paragraph b) of this example. and the rapamycin cassettes prepared as described in Example 2 were inserted into the *Pst*I/*Xba*I sites. The recipient plasmid was pCK7 described in Preparation A. The *in vivo* recombination technique resulted in the expression plasmid pKOS011-19 (eryDH/ER/KR4 → rapDH/ER/KR1) and pKOS011-21 (eryDH/ER/KR4 → rapDH/KR4). The junctions at which the *Pst*I and *Xba*I sites were introduced into DEBS in both vectors are as follows:

GAGCCCCAGCGGTACTGGCTGCAG rap cassette TCTAGAGCGGTGCAGGCGGCCCCG (SEQ ID NOS:32-33)--

Please replace the paragraph beginning at page 31, line 13, with the following rewritten paragraph:

--Approximately 1 kb regions flanking the eryKR6 domain were PCR amplified with the following primers (SEQ ID NOS:34-37) :

left flank	forward	5'-TTTGGATCCGTTTTCGTCTTCCCAGGTCAG
	reverse	5'-TTTCTGCAGCCAGTACCGCTGGGGCTCGAA
right flank	forward	5'-TTTTCTAGAGCGGTGCAGGCGGCCCCGGCG
	reverse	5'-AAAATGCATCTATGAATTCCCTCCGCCCA

Please replace the paragraph beginning at page 31, line 17, with the following rewritten paragraph:

--These fragments were then cloned into a pMAK705 derivative in which the multiple cloning site region was modified to accommodate the restriction sites of the fragments (i.e., *Bam*HI/*Pst*I for the left flank and *Xba*I/*Nsi*I for the right flank). Cassettes were then inserted into the *Pst*I/*Xba*I sites of the above plasmid to generate donor plasmids for the *in vivo* recombination protocol. The resulting *Pst*I and *Xba*I junctions engineered into DEBS are as follows:

GAACACCAGCGCTTCTGGCTGCAG rap cassette TCTAGAGACCGGCTCGCCGGTCGG (SEQ ID NOS:38-39)--

Please replace the paragraph beginning at page 32, line 11, with the following rewritten paragraph:

--d) Replacement of DEBS AT2. The DEBS AT activity from module 2 was excised after inserting restriction sites *Bam*HI and *Pst*I flanking the AT module 2 domain into pCK12 (Kao *et al. J Am Chem Soc* (1995) 112:9105-9106). After digestion with *Bam*HI/*Pst*I, the *Bgl*II/*Pst*I fragment containing rapAT2 was inserted. The resulting engineered DEBS/rapAT2 junction is as follows (*Bam*HI/*Bgl*II ligation - GGATCT; *Pst*I - CTGCAG):

AGTGCCTCCGACGGTGGATCT rapAT2 CTGCAGCCGGACCGCACCACCCCT (SEQ ID NOS:40-41)--

Please replace the paragraph beginning at page 32, line 28, with the following rewritten paragraph:

--A duplex oligonucleotide linker (ΔRdx) was designed to allow complete excision of reductive cycle domains. Two synthetic oligonucleotides (SEQ ID NOS:42-43)

5'-GCCGGACCGCACCACCCCTCGTGACGGAGAACCGGAGACGGAGAGCT-3'

3'-ACGTCGGCCTGGCGTGGTGGGGAGCACTGCCTCTTGGCCTCTGCCTCTCGAGATC-5'
PstI *XbaI*

were designed to generate *Pst*I- and *Xba*I-compatible ends upon hybridization. This duplex linker was ligated into the *Pst*I- and *Xba*I-sites of the recombination donor plasmid containing the appropriate left- and right-flanking regions of the reductive domain to be excised. The *in vivo* recombination technique of Example 3, paragraph a) was then used. The donor plasmid contained the duplex linker ΔRdx having a *Pst*I and *Xba*I compatible end ligated into the *Pst*I and *Xba*I sites of the plasmid modified to contain the left and right flanking regions of the reductive domain to be excised. The donor plasmids were recombined with recipient plasmid pCK7 to generate pKOS011-13 (eryKR6 → ΔRdx) and with recipient plasmid pCK13 to obtain pKOS005-4 (eryKR2 → ΔRdx). When transformed into *S. coelicolor* CH999, plasmid pKOS011-13 produced the polyketides 30a,b, 31a,b, 77a,b and 78a,b; in Figure 6 plasmid pKOS005-4 produced the polyketide 2a,b.--

Please replace the paragraph beginning at page 33, line 30, with the following rewritten paragraph:

--The 1+2+TE PKS in pCK12 contained a fusion of the carboxy-terminal end of the acyl carrier protein of module 2 (ACP-2) to the carboxy-terminal end of the acyl carrier protein of module 6 (ACP-6). Thus ACP-2 is essentially intact and is followed by the amino acid sequence naturally found between ACP-6 and the TE. Plasmid pCK12 contained *ery*A DNA originating from pS1 (Tuan, J. S. *et al. Gene* (1990) 90:21). pCK12 is identical to pCK7 (Kao *et al. Science* (1994), *supra*) except for a deletion between the carboxy-terminal ends of ACP-2 and ACP-6. The fusion occurs between residues L3455 of DEBS1 and Q2891 of DEBS3. An *Spe*I site is present between these two residues so that the DNA sequence at the fusion is CTCACTAGTCAG. (SEQ ID NO:44)--